



US009261482B1

(12) **United States Patent**
Henry et al.

(10) **Patent No.:** **US 9,261,482 B1**
(45) **Date of Patent:** **Feb. 16, 2016**

(54) **GRADIENT ELUTION MOVING BOUNDARY ELECTROPHORESIS FOR USE WITH COMPLEX SAMPLES AND DETECTION OF TOXINS**

(75) Inventors: **Alyssa Catharyn Henry**, Arlington, VA (US); **David Judson Ross**, Silver Spring, MD (US)

(73) Assignee: **Applied Research Associates, Inc.**, Albuquerque, NM (US)

(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 1024 days.

(21) Appl. No.: **12/761,515**

(22) Filed: **Apr. 16, 2010**

(51) **Int. Cl.**
G01N 27/447 (2006.01)
B01D 57/02 (2006.01)
G01N 30/28 (2006.01)

(52) **U.S. Cl.**
CPC **G01N 27/44713** (2013.01); **B01D 57/02** (2013.01); **G01N 27/4473** (2013.01); **G01N 2030/285** (2013.01)

(58) **Field of Classification Search**
CPC B01D 57/02; G01N 27/44713; G01N 27/447; G01N 27/4473; G01N 2030/285
USPC 204/450–470, 546–550, 600–621, 204/641–645
See application file for complete search history.

(56) **References Cited**

U.S. PATENT DOCUMENTS

5,098,826 A 3/1992 Wilkins et al.
5,482,608 A * 1/1996 Keely et al. 204/452

(Continued)

OTHER PUBLICATIONS

E. Strychalski, A. Henry, D. Ross, Microfluidic Analysis of Complex Samples with Minimal Sample Preparation Using Gradient Elution Moving Boundary Electrophoresis, *Anal. Chem.*, vol. 81, No. 24, p. 10201-10207, (2009).*

(Continued)

Primary Examiner — Luan Van

Assistant Examiner — Maris R Kessel

(74) *Attorney, Agent, or Firm* — Dinsmore & Shohl LLP

(57) **ABSTRACT**

Methods of detecting the presence of toxins in a sample using electrophoretic separations and of performing electrophoretic separation of complex samples are provided. The method of detecting the presence of toxins includes reacting a sample and a substrate with a signaling enzyme which converts the substrate to the product in a reaction medium, introducing a run buffer into a separation channel having an inlet end, selectively introducing at least one of the substrate and the product of the reaction medium into the inlet end of the separation channel, electrophoretically separating the substrate and the product, and determining the rate of conversion of the substrate to the product, wherein a change in the rate of conversion is indicative of the presence of toxins. The method of performing electrophoretic separations of complex samples having charged particulates and oppositely charged analytes comprising introducing a run buffer into a separation channel having an inlet end, selectively introducing the oppositely charged analytes in the complex sample into the separation channel, and electrophoretically separating the charged particulates and the oppositely charged analytes. Additionally, a device for varying with respect to time the bulk flow of a fluid in a separation channel of an electrophoretic device having a buffer reservoir in fluid contact with the separation channel is provided. The device includes a pressure sensor in fluid contact with a buffer reservoir, a high pressure reservoir in selective fluidic communication with the buffer reservoir, a low pressure reservoir in selective fluidic communication with the buffer reservoir and in fluidic communication with the high pressure reservoir, and a pumping device for pumping a gas from the low pressure reservoir to the high pressure reservoir.

20 Claims, 18 Drawing Sheets

